PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference MP020096-WO	**FOR FURTHER ACTION**	See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416)
International application No. PCT/GB 03/02420	International filing date *(day/month/year)* 02.06.2003	Priority date *(day/month/year)* 31.05.2002
International Patent Classification (IPC) or both national classification and IPC G01N33/68		
Applicant SHIMADZU RESEARCH LABORATORY (EUROPE) LIM.., et al		

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 6 sheets, including this cover sheet.

   ☐ This report is also accompanied by ANNEXES, i.e. sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

   These annexes consist of a total of sheets.

3. This report contains indications relating to the following items:

   - I ☒ Basis of the opinion
   - II ☐ Priority
   - III ☒ Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
   - IV ☐ Lack of unity of invention
   - V ☒ Reasoned statement under Rule 66.2(a)(ii) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
   - VI ☐ Certain documents cited
   - VII ☐ Certain defects in the international application
   - VIII ☐ Certain observations on the international application

Date of submission of the demand	Date of completion of this report
17.12.2003	09.12.2004
Name and mailing address of the international preliminary examining authority: European Patent Office D-80298 Munich Tel. +49 89 2399 - 0 Tx: 523656 epmu d Fax: +49 89 2399 - 4465	Authorized Officer Lopez García, F Telephone No. +49 89 2399-2171

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No. PCT/GB 03/02420

I. Basis of the report

1. With regard to the **elements** of the international application *(Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report since they do not contain amendments (Rules 70.16 and 70.17))*:

Description, Pages

1-27 as originally filed

Claims, Numbers

1-27 as originally filed

Drawings, Sheets

1/13-13/13 as originally filed

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.

   These elements were available or furnished to this Authority in the following language: , which is:

   ☐ the language of a translation furnished for the purposes of the international search (under Rule 23.1(b)).

   ☐ the language of publication of the international application (under Rule 48.3(b)).

   ☐ the language of a translation furnished for the purposes of international preliminary examination (under Rule 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

   ☐ contained in the international application in written form.

   ☐ filed together with the international application in computer readable form.

   ☐ furnished subsequently to this Authority in written form.

   ☐ furnished subsequently to this Authority in computer readable form.

   ☐ The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.

   ☐ The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. The amendments have resulted in the cancellation of:

   ☐ the description, pages:

   ☐ the claims, Nos.:

   ☐ the drawings, sheets:

5. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed (Rule 70.2(c)).

   (Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report.)

6. Additional observations, if necessary:

III. Non-establishment of opinion with regard to novelty, inventive step and industrial applicability

1. The questions whether the claimed invention appears to be novel, to involve an inventive step (to be non-obvious), or to be industrially applicable have not been examined in respect of:

   ☐ the entire international application,

   ☒ claims Nos. 1-22, 24-27 (all partially)

   because:

   ☐ the said international application, or the said claims Nos. relate to the following subject matter which does not require an international preliminary examination (specify):

   ☐ the description, claims or drawings *(indicate particular elements below)* or said claims Nos. are so unclear that no meaningful opinion could be formed *(specify)*:

   ☐ the claims, or said claims Nos. are so inadequately supported by the description that no meaningful opinion could be formed.

   ☒ no international search report has been established for the said claims Nos. 1-22-24-27 (all partially)

2. A meaningful international preliminary examination cannot be carried out due to the failure of the nucleotide and/or amino acid sequence listing to comply with the standard provided for in Annex C of the Administrative Instructions:

   ☐ the written form has not been furnished or does not comply with the Standard.

   ☐ the computer readable form has not been furnished or does not comply with the Standard.

V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Yes:	Claims	1-27 (partially)
	No:	Claims	
Inventive step (IS)	Yes:	Claims	1-27 (partially)
	No:	Claims	
Industrial applicability (IA)	Yes:	Claims	1-27 (partially)
	No:	Claims	

2. Citations and explanations

   see separate sheet

Re Item III

Non-establishment of opinion with regard to novelty, inventive step and industrial applicability

1. The applicant did not reply to the invitation to pay additional search fees. This opinion will be established exclusively for the searched subjet-matter (Rule 66.1(e) PCT).

Thus, no opinion will be issued for inventions 2-5 cited by the ISA, which correspond to claims 1-22, 24-27 (all partially).

Re Item V

Reasoned statement with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

2. This opinion relates exclusivelty to invention 1 cited by the ISA, namely to the reagent "target A" and its use in quantification of protein in complex mixtures, which correspond to claims 1-27 (all partially).

3. Reference is made to the following documents:

D1: CAGNEY GERARD ET AL: "De novo peptide sequencing and quantitative profiling of complex protein mixtures using mass-coded abundance tagging" NATURE BIOTECHNOLOGY, vol. 20, no. 2, February 2002 (2002-02), pages 163-170, XP001155365 ISSN: 1087-0156

D2: BRANCIA F L ET AL: "A combination of chemical derivatisation and improved bioinformatic tools optimises protein identification for proteomics." ELECTROPHORESIS. GERMANY FEB 2001, vol. 22, no. 3, February 2001 (2001-02), pages 552-559, XP002257401 ISSN: 0173-0835

D3: DATABASE CROSSFIRE BEILSTEIN [Online] BEILSTEIN INSTITUT ZUR FOERDERUNG DER WISSENSCHAFTEN, FRANKFURT AM MAIN, DE; XP002257403 Database accession no. 1753773

D4: DATABASE CROSSFIRE BEILSTEIN [Online] INSTITUT ZUR FOERDERUNG DER WISSENCHAFTEN, FRANKFURT AM MAIN, DE; XP002257821 Database accession no. 2096913

D5: DATABASE CROSSFIRE BEISLTEIN [Online] INSTITUT ZUR FOERDERUNG DER WISSENCHAFTEN, FRANKFURT AM MAIN, DE; XP002258054 Database accession no. 3337097

D6: DATABASE CROSSFIRE BEILSTEIN [Online] INSTITUT ZUR FOERDERUNG DER WISSENCHAFTEN, FRANKFURT AM MAIN, DE; XP002258055 Database accession no. 1489771

D7: WO 00/11208 A (UNIV WASHINGTON) 2 March 2000 (2000-03-02)

D8: GYGI S P ET AL: "QUANTITATIVE ANALYSIS OF COMPLEX PROTEIN MIXTURES USING ISOTOPE-CODED AFFINITY TAGS" NATURE BIOTECHNOLOGY, NATURE PUBLISHING, US, vol. 17, no. 10, October 1999 (1999-10), pages 994-999, XP001010578 ISSN: 1087-0156

4. The present subject-matter relates to the analysis of proteins in complex mixtures by guanidination of Lys residues with the target A (see Fig. 10).

5. Novelty (Art. 33(2) PCT).

(D1) discloses the guanidination with O-methylisourea of Lys residues to quantify proteins in complex mixtures (see Abstract and Fig. 1A).

(D2) discloses the guanidination of Lys from proteins in a complex mixture with o-methylisourea (see abstract and p. 553, col. 1, paragraph 2).

(D3), (D4), (D5), (D6) disclose compounds acording to claim 22 (see the whole document).

(D7) and (D8) disclose the ICAT method and the derivatization of Cys residues.

Therefore, the compound "Target A" of figure 10 of the present application is novel

6. Inventive step (Art. 33(3) PCT).

D1 is considered the closest prior art. D1 is directed to the derivatization of Lys residues with o-methylisourea.

The present application differs from D1 is that the reagent used is an o-methylisourea derivative comprising biotin.

The technical problem is the provision of alternative compounds for the derivatization of Lys.

The technical problem is solved by the present reagent A as shown in the examples (Fig. 11).

The prior art neither discloses nor suggests the present target A, let alone its use in the derivatization of Lys. Furthermore, as indicated in the examples said target A, the results obtained are quantitative and there is not derivatization of the N-terminal amino group (p. 26, second paragraph).

On the other hand, the target A of the present application is not obvious by combination of D1 and D7 (or D8). These documents (D7-D8) are directed to the derivatization of Cys residues with a reagent comprising biotin and a linker, whereas the present application is directed to the derivatization of Lys residues. The compounds are therefore not interchangeable. Moreover, the linker of D7 is different from that used of target A. Even in the case that the skilled person would the prompted to produce biotinylated derivatives of the reagent of D1, the present compound is not suggested.

7. Industrial applicability (Art. 33(4) PCT).

The subject-matter of claims 1-27 (partially) is industrially applicable.

Certain defects in the international application

8. Contrary to the requirements of Rule 5.1(a)(ii) PCT, the relevant background art disclosed in the documents D1 and D2 is not mentioned in the description, nor are these documents identified therein.

Certain observations on the international application

9. The claims are not clear (Art. 6 PCT), since the meaning of X, R or L is not given in the claims. The definition given at p. 11 for X, R and L is not found in the claims.

10. Page 19 refers to Fig. 10, compound B. However, the compounds of figure 10 are named as target A-E. Moreover, target B is not a biotin derivative.

11. The target A, as indicated in figure 10, does not correspond to the compound obtained in the synthesis described at p. 19. Biotin has not two methyl groups in the ring.